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Docket No. SPO-112  
Serial No. 09/762,842Remarks

Claims 1-14 are pending in the subject application. By this Amendment, the applicants have amended claims 2, 5, 8 and 10. Support for the amendments set forth herein can be found throughout the subject specification. Accordingly, claims 1-14 are currently before the Examiner. Favorable consideration of the pending claims is respectfully requested.

The amendments set forth herein have been made to more clearly delineate the claimed subject matter and to expedite prosecution. These amendments should not be construed as an indication of the applicant's agreement with, or acquiescence in, the outstanding rejections.

Initially, the applicants wish to acknowledge, and express their gratitude, for the Examiner's indication of allowable subject matter. The claims have been amended herein to more clearly focus on the allowable subject matter and address the other remaining issues as discussed below.

Claims 1-4 and 10 have been rejected under 35 USC §112, second paragraph, as being indefinite. The applicants respectfully traverse this grounds for rejection because the claims, as presently amended, clearly and unambiguously set forth the metes and bounds of the claimed subject matter.

The Office Action indicates that the recitation of "5' and 3' sequences within the AOX1 gene" renders the claims indefinite because it is unclear to which part of the AOX1 gene the applicants are referring. Initially, the applicants note that only claims 2 and 10 refer to 5' and 3' AOX1 sequences. Accordingly, the instant rejection does not appear appropriate for claims 1, 3, and 4. In any event, the applicants respectfully submit that all the claims, as presently amended, fully satisfy the requirements of 35 U.S.C. §112, second paragraph. Of course, the test for indefiniteness is whether one of ordinary skill in the art could discern and understand the metes and bounds of the claim, when read in light of the specification and in the context of what is known in the art. Thus, claim language should not be analyzed in a vacuum but should instead be interpreted in light of the specification, the teachings of the art, and the reasonable interpretation given by one of ordinary skill in the art.

In this case, in the context of this art, it is clear from the specification that the claimed sequences must comprise the 5' and 3' ends of the entire AOX1 gene sequence in order to perform the claimed function - namely, site specific homologous recombination to the yeast chromosome. Accordingly, in the present context, the term "AOX1 gene" necessarily refers to the complete

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nucleotide sequence of the gene, including both coding and non-coding regions. Accordingly, when the claimed terminology is interpreted in light of the guidance set forth in the specification and in the context of what is known in the art, the meaning of the claims becomes quite clear.

Nevertheless, to expedite prosecution, the applicants have amended the relevant claims to clarify that, consistent with the teachings of the specification and the art, the sequences at issue comprise the 5' and 3' end sequences of the AOX1 gene (i.e., complete nucleotide sequence including untranslated, non-coding regions), wherein these sequences facilitate site-specific homologous recombination to a methylotrophic yeast chromosomal DNA. Specifically, by flanking a vector DNA with sequences taken from the 5' and 3' ends of the AOX1 gene, one can induce the integration of the vector DNA into the host cell (i.e., yeast) chromosome through site-specific homologous recombination.

In view of the clarity of these claims as amended, the applicants respectfully request reconsideration and withdrawal of the rejection under 35 U.S.C. §112, second paragraph.

Claims 1-5, 8 and 10-12 have been rejected under 35 USC §103(a) as being unpatentable over Davis *et al.* (WO 92/13951), in view of Tomomura *et al.* (1990, JBC, Vol. 265, No. 18, pp. 10765-10770) and Li *et al.* (1990, Science 250:1690-1694).

The Office Action indicates that the applicants' arguments of unexpected results are unpersuasive because the claims do not recite limitations of increased production or producing MK protein at a specific pH. In this regard, claim 9, which recites the noted pH limitation, has been indicated to contain allowable subject matter but has been objected to as depending from a rejected base claim. Accordingly, consistent with the Examiner's indication of allowable subject matter, claim 8 has been amended herein to refer to the specific pH.

Also, claim 5 has been amended herein to specifically state that the claimed vector is for "enhanced" expression of an intact MK family protein. On the issue of unexpected results, it is well established that objective evidence of non-obviousness should be commensurate in scope with the claim in order to be persuasive. However, there is no requirement for the unexpected result or feature to itself be an element of the claim. Rather, all that is required is that the experimental evidence *reflect* the claimed invention. Thus, once novelty has been established (as it has been in the present

case), the unexpected results must simply be shown to be attributed to the novel subject matter in order to overcome a *prima facie* case of obviousness.

In this case, superior results are directly attributable to the unique combination of elements claimed, e.g., the use of the  $\alpha 1$  secretion signal from *S. cerevisiae* to induce secretory expression of an MK protein. Specifically, the applicants have shown that using the secretion signal unique to MK results in low yield and the predominance of sugar-bound MK (i.e., the expression of intact MK protein is low). Conversely, ligation of the MK protein cDNA immediately downstream of the prepro-sequence of  $\alpha 1$  factor gene of *S. cerevisiae* under the regulation of the AOX1 promoter from *P. pastoris* results in a marked increase in expression of intact MK protein. See specification at p. 3, lines 12-16. For example, as discussed in Example 3, the expression level of MK produced using the  $\alpha 1$  factor as the secretion signal (pPIC9K-4AhMK) was 100 times that detected using the PHO1 secretion signal (PHILS1-3AhMK) and 3 to 5 times that detected using the secretion signal unique to MK (PHILD4-hMK). Thus, the applicants' data demonstrates the advantageous and unexpected benefits of using the instantly claimed secretion signal.

Contrary to the current invention, Davis et al. suggest that the secretion signal may be selected either from the *S. cerevisiae* AMF (i.e., the  $\alpha 1$  factor) or the native protein secretion signal (see Abstract, lines 3-4 as well as p. 7, lines 9-20; p. 11, lines 29-34; and p. 13, lines 3-29). Thus, the Davis reference fails to recognize or even suggest the criticality of using the claimed secretion signal sequence to achieve unexpected high levels of expression of the MK protein.

Thus, the unexpected results are commensurate with the scope of the claims and sufficient to rebut a *prima facie* case of obviousness. Accordingly, the applicants respectfully request reconsideration and withdrawal of the rejection of claims 1-5, 8, and 10-12 under 35 U.S.C. § 103.

Finally, with regard to the drawings in the subject application, the applicants are attaching for the Examiner's convenience evidence that the drawings were, in fact, submitted (and received by the Patent Office) at the time of filing. This evidence includes the Notice of Acceptance of Application under 35 U.S.C. § 371, which acknowledges the receipt of the International Application (IA) and the English translation of the IA. It cannot be disputed that the IA contained the drawings. The applicants are further submitting a copy of the official Filing Receipt, which specifically acknowledges 17 drawings. The applicants appreciate that the drawings may have inadvertently

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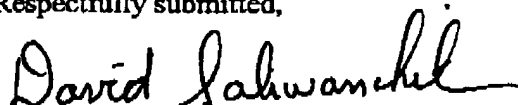
been separated from the file and, therefore, have provided new copies of the drawings. However, it would be an unfair and unnecessary burden for the applicants to be required to petition to the Patent Office to have these drawings entered when the record so clearly indicates that they were received by the Patent Office at the time of filing.

In view of the foregoing remarks and amendments to the claims, the applicants believe that the currently pending claims are in condition for allowance, and such action is respectfully requested.

The Commissioner is hereby authorized to charge any fees under 37 CFR §§1.16 or 1.17 as required by this paper to Deposit Account No. 19-0065.

The applicants invite the Examiner to call the undersigned if clarification is needed on any of this response, or if the Examiner believes a telephonic interview would expedite the prosecution of the subject application to completion.

Respectfully submitted,



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